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File: USPT

Sep 28, 1999

US-PAT-NO: 5958672

DOCUMENT-IDENTIFIER: US 5958672 A

TITLE: Protein activity screening of clones having DNA from uncultivated microorganisms

DATE-ISSUED: September 28, 1999

INVENTOR-INFORMATION:

NAME	CITY	STATE	ZIP CODE	COUNTRY
Short; Jay M.	Encinitas	CA		

US-CL-CURRENT: 435/4; 435/183, 435/69.1, 536/23.1, 536/23.2

CLAIMS:

What is claimed is:

1. A method for identifying a protein activity of interest comprising:

culturing a gene expression library comprising a pool of expression constructs, each expression construct comprising a vector containing one or more cDNA or genomic DNA fragments, wherein the cDNA or genomic DNA fragments in the pool of expression constructs are derived from a plurality of species of donor organisms, and wherein the cDNA or genomic DNA fragments are each operably-associated with one or more regulatory regions that drives expression of genes encoded by the cDNA or genomic DNA fragments in an appropriate host organism; and

detecting the protein activity encoded by the cDNA or genomic DNA fragments.

2. The method of claim 1, wherein the protein activity is an enzymatic activity.

3. The method of claim 2, wherein the enzymatic activity is selected from the group consisting of oxidoreductase, transferase, hydrolase, lyase, isomerase, and ligase activity.

4. The method of claim 1, wherein the donor organisms are microorganisms.

5. The method of claim 4, wherein the microorganisms are derived from an environmental sample.

6. The method of claim 4, wherein the microorganisms are a mixed population of uncultured organisms.

7. The method of claim 1, wherein the DNA fragment comprises one or more operons, or portions thereof.

8. The method of claim 7, wherein the operon or portions thereof encodes a complete or partial metabolic pathway.

9. A method for identifying a protein activity of interest comprising:

culturing a gene expression library, comprising a pool of expression constructs,

each expression construct comprising a vector containing one or more cDNA or genomic DNA fragments, wherein the cDNA or genomic DNA fragments in the pool of expression constructs are derived from a plurality of species of donor microorganisms, and wherein the cDNA or genomic DNA fragments are each operably-associated with one or more regulatory regions that drives expression of genes encoded by the cDNA or genomic DNA fragments in an appropriate host organism; and

detecting the protein activity encoded by the cDNA or genomic DNA fragments.

10. The method of claim 9, wherein the protein activity is an enzymatic activity.

11. The method of claim 10, wherein the enzymatic activity is selected from the group consisting of oxidoreductase, transferase, hydrolase, lyase, isomerase, and ligase activity.

12. The method of claim 9, wherein the microorganisms are derived from an environmental sample.

13. The method of claim 9, wherein the microorganisms are a mixed population of uncultured organisms.

14. The method of claim 9, wherein the DNA fragment comprises one or more operons, or portions thereof.

15. The method of claim 14, wherein the operon or portions thereof encodes a complete or partial metabolic pathway.

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<u>L6</u>	(enzym\$3 and activ\$3 and clon\$3 and organis\$3 and librar\$3 and screen\$3).clm	16463	<u>L6</u>
<u>L5</u>	(enzym\$3 and clon\$3 and organis\$3 and librar\$3 and screen\$3).clm	16855	<u>L5</u>
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<u>L2</u>	uncultivat\$3 and clon\$3 and organis\$3 and librar\$3	89	<u>L2</u>
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